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Feasibility of phytoremediation of common soil and groundwater pollutants

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timbre

Tailored Improvement of
Brownfield Regeneration
in Europe

Feasibility of phytoremediation of common soil and groundwater pollutants

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Feasibility of phytoremediation of common soil and groundwater pollutants

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1. Summary

This report is the Deliverable D4.3 and was done within the Timbre project WP4. It introduces into the various clean-up techniques that apply plants, evaluates the feasibility of phytoremediation of common soil and groundwater pollutants, and the knowledge collected for this purpose was applied to the two Timbre sites: Hunedoara (Romania) and Szprotawa (Poland). Phytoremediation is the technique to clean up (remediate) contaminated sites using plants, typically trees. The principles of the data were detailed, with focus on obstacles (phytotoxicity) and factors stimulating success (degradation). Application schemes were established. Quantitative data was collected and presented in tables. Kinetic models were established for later application at the test sites.

The feasibility of phytoremediation at the Timbre sites was evaluated. The former steel works site Hunedoara has occasionally extremely high concentrations of toxic elements and heavy metals in top soil. At these places, phytotoxicity is likely to occur and would inhibit or distort phytoremediation efforts. Moreover, the pH at site is rather high (7.65 to 8.97), which leads to very strong sorption (K_d) of several heavy metals (in particular, nickel and lead). These high K_d values limit uptake of heavy metals, and calculated clean-up times are unrealistic (millions of years). In conclusion, phytoremediation is no option for Hunedoara.

At the Polish site Szprotawa, which is a former military airbase, jet fuel and BTEX in >2 m depth in soil and groundwater is the major pollution. The toxicity of gasoline to trees has been studied both in lab and field and is quite well known. High ROZ fuel is lethal at concentrations of about 1,000 mg/kg soil. Concentrations at Szprotawa are often higher (up to 11,000 mg/kg), but the peak is deep below surface (2 m depth). Thus, tree growth is possible but may be inhibited at longer periods of drought (when trees root deeper to get water supply). Vividly growing pioneer vegetation has been observed at site, confirming this finding. Hydrocarbons are rapidly degraded under aerobic conditions, and the immediate reaction model with diffusion of oxygen as limiting factor was applied. The calculations show that jet fuel and BTEX in top soil will degrade quickly. Complete degradation under optimal conditions would occur in a bit more than a decade. Experience from real sites teaches, however, that hydrocarbons residing below the groundwater level will persist for longer periods. Taking all together, phytoremediation is a good option to support the naturally occurring degradation of contamination at the site.

2. Introduction

The **TIMBRE project** shall overcome existing barriers to brownfield regeneration by developing and providing customised problem- and target-oriented packages of approaches, technologies and tools. As a unique asset, these packages shall deliberately include the cultural and administrative characteristics and their regionally distinctive features. By providing a customisable toolbox specifically addressing the diverse processes that have to be dealt with during the course of a regeneration project, end-users shall be enabled to find best practice based solutions. Improvement of existing means to support brownfield regeneration shall be further accomplished by filling methodological core topics such as intelligent remediation in terms of technological advancements with regard to phytoremediation and partial source removal technologies. Find further information on the project on <http://www.timbre-project.eu>.

In **Work Package 4** (WP4) strategies and technologies for integrated site characterisation and remediation are investigated. One of the main tasks of WP4 is to assess the feasibility of vegetation as a remediation- and risk reducing tool for typical pollutants in the subsurface.

Using vegetation as a remediation tool for contaminated sites or as a mean to lower contaminant risk is called phytoremediation. Phytoremediation can be implemented with many different plant species; however, the studies presented in this report are based on trees, mainly willows and poplars.

The report presents an end users toolbox, containing a decision flow chart for phytoremediation, two mass balance models, with and without degradation, for assessing the timescale of phytoremediation for a given site and phytotoxicological data for common soil and groundwater pollutants. To demonstrate the applicability of the models they have been applied for the TIMBRE sites of Hunedoara, Romania, and Szprotawa, Poland.

The purpose of this report is to describe the phytoremediation method and to evaluate the feasibility of the method for soil and groundwater pollutants. Further, the report aims to provide the end user with adaptable tools for assessing the potential of phytoremediation at local case sites.

3. Phytoremediation

The phytoremediation technology arose in the nineties and early millennium. The expectancies were high as research seemed promising (Pulford and Watson, 2002, Salt et al., 1998 and Pradhan et al., 1998). However, phytoremediation is still not widely applied. This may be a consequence of the too promising expectations and the tendency to oversell the technology.

Phytoremediation covers different remediation strategies, processes and purposes. These occur simultaneously and do not exclude or prohibit each other (Trapp and Karlson, 2001). Despite this, the different processes may or may not be desired in specific cases.

3.1 Definition of phytoremediation

Phytoremediation is a technique for remediating polluted soils by use of plants (Trapp and Karlson 2001). Pollutants can both be inorganic and organic chemicals. Phytoremediation has several benefits: It is inexpensive, it seems effective, it is in situ, and it is "green" (Flathman and Lanza, 1998; Schnoor et al., 1995). A special advantage of phytoremediation compared to other techniques is that soil functioning is maintained and life in soil is reactivated.

Trees, grasses, herbs, and associated fungi and microorganisms are being used for cleaning polluted sites. Phytoremediation has a good image and is often, but not always, more cost-effective than competing techniques. But successful phytoremediation takes time, sometimes more than one decade, which makes it difficult to use this technique in urban areas where brownfields and other contaminated sites shall be used for new buildings and other usage. The use of phytoremediation may provide a useful tool for reaching the desired remediation goals or mitigating the environmental problems as well as functioning as a risk-reducing treatment, but the processes are slow and the remediation time measures in decades or centuries.

3.2 Implementing phytoremediation – Purposes and processes

The act of implementing phytoremediation is simple compared to other remediation technologies. Firstly, the contamination is delineated by conventional methods. Secondly, dependent on the remediation strategy and the local conditions, appropriate plant species are planted out in the area of concern. Best success is when planting in spring, when sufficient water is available for immature plants. Either before or after transplanting of plants proper monitoring of the site needs to be established to follow the remediation progress. Preferably, boreholes and installations for delineation of the polluted site can be reused for monitoring purposes. In some cases addition of nutrition may be desired in order to make the plants thrive. This depends highly on the soil properties of the site and on the remediation strategy applied. E.g. for phytoextraction, where biomass is removed from the site, inhibition of plant growth may occur because of lack of nutrients.

Several phytoremediation techniques have been described in Trapp and Karlson (2001), and we cite from there (shortened): Phytoextraction, rhizofiltration, phytostabilization, rhizodegradation, phytodegradation, phytovolatilization, hydraulic control, vegetation cover and buffer stripes.

Phytoextraction

Phytoextraction signifies the uptake, translocation and accumulation of pollutants in plants. Harvest products, which concentrate the pollutants, may be used or disposed of. The technique is preferably used for heavy metals that cannot be degraded in the root zone and are non-volatile.

Rhizofiltration

Rhizofiltration is the sorption of contaminants to roots or other plant parts, or the precipitation in the root zone. E.g., heavy metals or lipophilic compounds can be extracted from water by this technique.

Phytostabilization

Phytostabilization is the immobilization of compounds in soil, or the stabilizing of soil itself to prevent erosion. In the first case, pollutants are transferred from a soluble form into a non-soluble form by the redox milieu in the root zone.

Rhizo- und phytodegradation

Phytodegradation is the degradation of pollutants by plants. Rhizodegradation is the degradation of contaminants in the root zone, either due to microbial activity or by roots, or by both. In the root zone, several processes accelerate degradation of some compounds (see above). Phyto- and rhizodegradation are frequently used for the remediation of organic contaminations, among them petroleum, PAH, BTEX, TNT, chlorinated solvents and pesticides (EPA 2000).

"Pump and Tree"

One of the common remediation techniques for groundwater pollution is "pump and treat", pumping of water with subsequent technical cleaning (stripping, adsorption, bioreactor etc.). A new idea is to use pumped water during times of negative water balance (summer) for irrigation, e.g. of forests. One ha Salix stand can transpire up to 3000 m³ water in July (Larcher 1995). Forest soils have a high metabolic capacity and might degrade many compounds quickly. At least a part of the pumped water should be treatable by trees. Chemicals to be treated by this method are organic solvents (trichloroethene), MTBE, petroleum products, nutrients and perhaps some others.

Land farming

Another treatment method based on root zone degradation is "land farming". The method is e.g. used for oil-polluted sludge: The sludge is ploughed into topsoil, the field is fertilized and alfalfa or grass (usually rye) is sowed out. In the rooted, aerated and fertilized topsoil, oil is degraded quickly.

Phytovolatilization

In phytovolatilization plants are used for extraction and sub-sequent out-gassing of compounds from soil. The process was shown to be relevant for chlorobenzene (Baeder-Bederski et al., 1999), trichloroethene (Orchard et al. 2000) and other volatile compounds (Burken and Schnoor, 1998), but also for organically bound mercury (EPA 2000). The technique is relevant for all compounds that are quickly translocated ($\log KOW < 3.5$) and have a high vapor pressure or a high Henry's Law constant ($KAW \gg 10^{-5}$ L/L). Gassing out only removes the pollution problem from one environmental medium to the other and is therefore seen as an unwanted by-process. However, translocation, and therefore volatilization, occur mainly when the sun is shining - this makes photolysis likely.

Hydraulic control

Very often, water is pumped to prevent leaching or movement of pollutants. Hydraulic control may be done partly or completely by trees or other plants, saving costs. In principle, this method can be applied for all contaminants, as long as the plants have no contact with the toxicants. The harvest products can be used without limitation. The main purpose of this technique is to combine it with mechanical pumping to reduce energy consumption and costs.

Vegetation cover

Waste deposits are frequently planted with grass, simply because it looks better, but also to avoid erosion. Some other aspects are beneficial: The infiltration of water is reduced, small amounts of escaping gas are adsorbed by plants (higher amounts may be toxic, EPA 2000). In only a few cases, planting trees on landfills is accepted by authorities, although trees transpire more and are esthetically even more pleasing than grasses. But it is feared that roots could go through the coverage and damage it. According to Dobson and Moffat (1995), these fears have no basis.

Buffer stripes

The planting of trees, e.g. poplars, along rivers is surely nothing new. It is also known that these stripes provide barriers for the run-off of nutrients from fields. Recently it was found that poplars can reduce the herbicidal load (atrazine) of creeks (Burken and Schnoor 1996).

Schnoor and co-workers evaluated applicability of phytoremediation (Schnoor et al., 1995, Schnoor, 1997). They found that the technique is most successful when the topsoil is polluted with chemicals being either degraded in the rhizosphere or effectively taken up by plants. For too high pollutant concentrations, toxic effects may occur, and phytoremediation therefore is restricted to lower to medium contamination levels. Thus, two key criteria for the success of phytoremediation projects are phytotoxicity (survival of plants) and biodegradability of contamination.

4. Decision tool

As described in several publications (e.g., Schnoor 1997, Trapp and Karlson 2001), the processes of phytoremediation are complex. This makes it strenuous to assess the applicability and feasibility of the method. To clarify the options available within the phytoremediation technology, their feasibility and ease the decision processes of decision makers, a phytoremediation flow chart has been constructed. The chart is divided into an A and a B part. Part A concerns the applicability of phytoremediation and deals with data which have to be collected, various tests may have to be performed if no data is available in the literature, in order to make the right decisions. The knowledge obtained in part A is applied in flow chart B, which specifies the expected dominating remediation processes.

4.1 Flow chart

The flow chart is based on earlier decision trees (ITRC 1999, Trapp 2000) and has been refined for the TIMBRE needs.

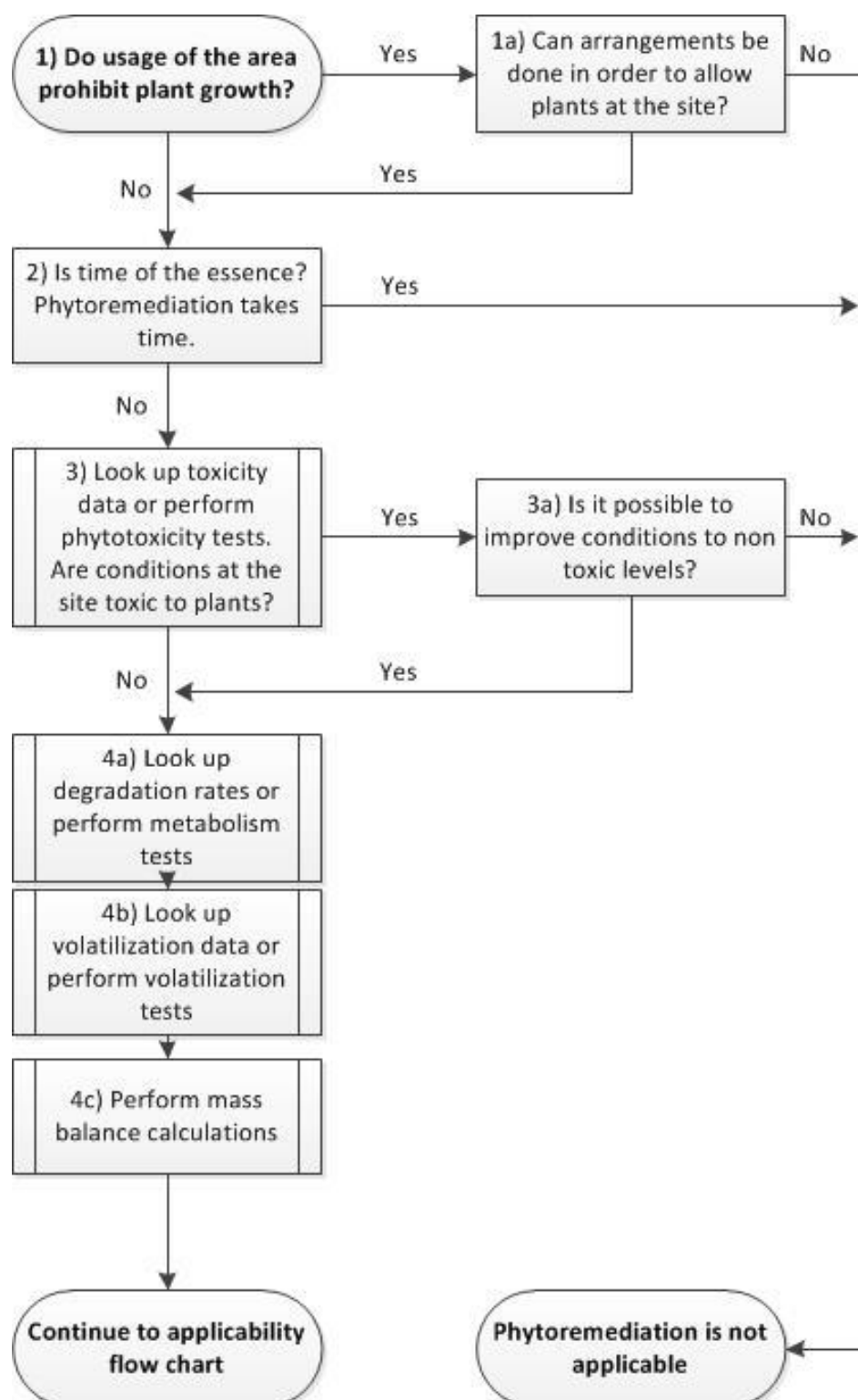


Figure 4.1A Flow chart A for applicability of phytoremediation.

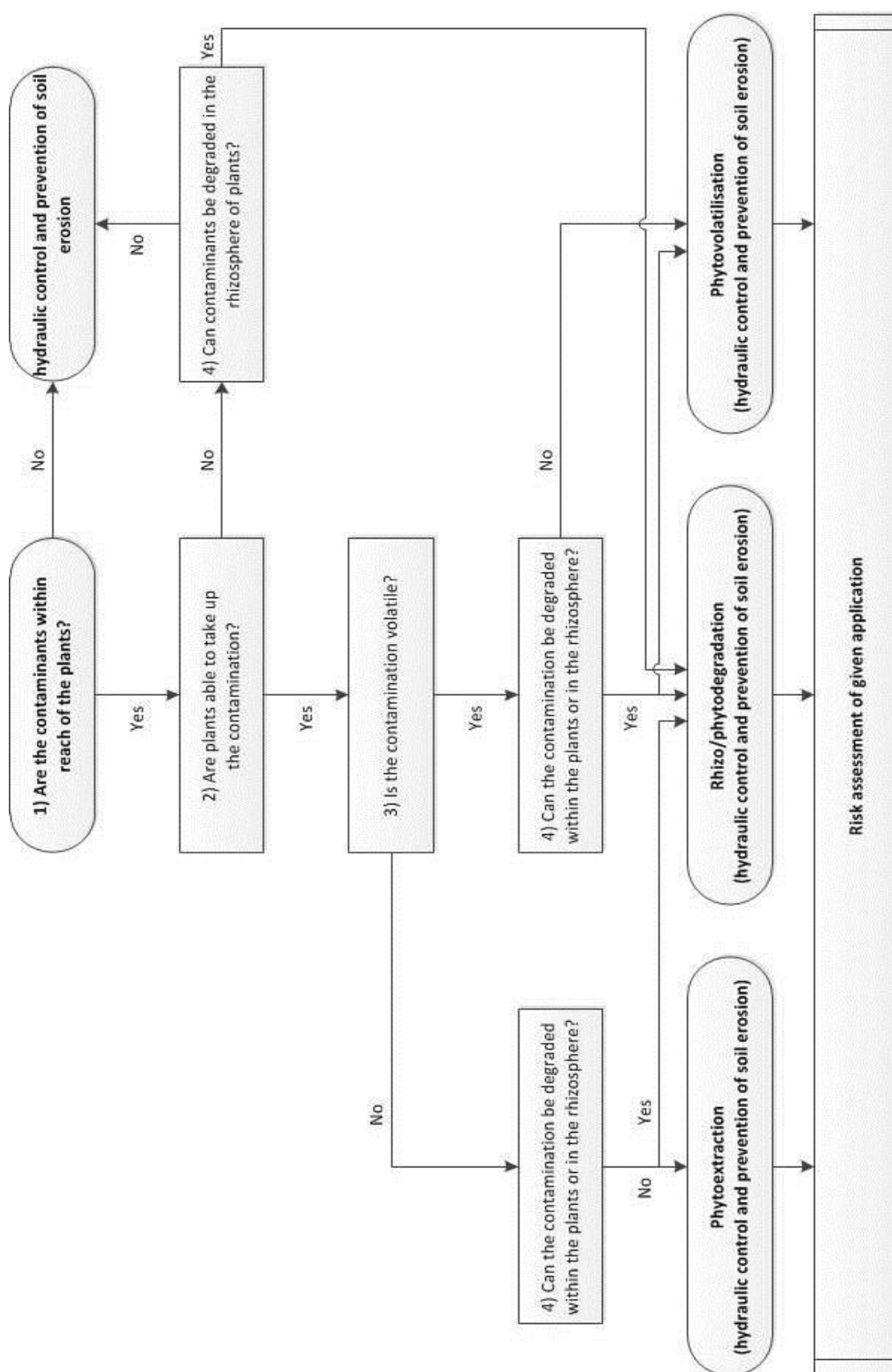


Figure 4.1B Flow chart B for applicability of phytoremediation techniques

4.2 Flow chart elaboration

Flow chart A: Applicability of phytoremediation

1: Does current or future usage of the area prohibit plant growth?

Will there be any activity at the application site which is or may become in conflict with plant growth in the area? The area might be used for different purposes during treatment and it is crucial that plants are allowed to be properly established.

1a: Can arrangements be done in order to allow plants at the site?

If plants cannot grow at the site phytoremediation will not be an option. However, if phytoremediation is still the desired treatment technology something will have to be done to protect or enhance the conditions for the plants.

2: Is time of the essence? Phytoremediation takes time.

Is it acceptable to let the site of concern undergo treatment for decades? In some cases treatment will have to be done for even hundreds of thousands of years dependent of the soil properties and the physico-chemical properties of the pollutants. At a later state the remediation time should be assessed by mass balance models, see point 4c, but as an initial action it is pivotal to consider whether a long remediation time is problematic.

3: Look up toxicity data or perform phytotoxicity tests. Are conditions at the site toxic to plants?

If plants die at the remediation area due to phytotoxic effects of the pollutant, phytoremediation will not be applicable. To assess this issue it is critical to know about phytotoxicity of the compounds present. This report includes phytotoxicological data for the most common soil and groundwater pollutants, see chapter 5. If no data is available it is recommended to perform standardized phytotoxicity test. For guidance in a suitable test, the reader is referred to Trapp et al. (2000).

It is important to assess whether the site is suited for plant growth or not. It may seem obvious but plants do not thrive under all conditions – temperature, humidity, precipitation, soil conditions, nutrients, salinity of the aquifer etc. It is important to make use of natural occurring plants also not to introduce foreign possible invasive species.

3a: Is it possible to improve conditions to non-toxic levels?

Plants will not grow at the target area and something needs to be done. If the toxicity is due to soil concentrations of contaminants, removal of hotspot(s) may be the best option. The option to use more resistant plants, which can cope with the high concentrations, is also worth to consider.

If the plant growth is inhibited by other factors these should be addressed. Fertilizing or irrigating the plants might solve the issue.

4a: Look up degradation rates or perform metabolism tests.

Plants and their associated rhizospheric microorganisms have been shown capable of degrading several types of pollutants (Barac et al., 2009, Doty et al., 2003, Newman et al., 1997, Newman et al., 1999 and Shang et al., 2001). To determine the fate of the contaminants it is vital to know if they are degradable or not. Data on degradation rates and degradability for various compounds may be found in literature. If not, it is recommended to obtain the required knowledge by performing metabolism studies for both plants and microorganisms. An example of a microbial degradation study of BTEX is provided by Chapelle et al. (1996).

4b: Look up volatilization data or perform volatilization tests.

If the plants can take up volatile components, they will be able to volatilize them to the atmosphere. Data for volatilization of the present contamination should be looked up in the literature. If nothing is available tests for determination of Henry's law constant should be performed. Phytovolatilization should be possible for compounds with Henry's law constant clearly higher than water ($> 6 \times 10^{-6}$ L/L, Trapp and Matthies 1998).

4c: Perform mass balance calculations.

The time needed for treatment should be assessed by use of models. This will as a minimum give an indication of the order of magnitude of the timeframe. This report presents two mass balance models for assessing the remediation time, see chapter 7.

Flow chart B: Applicability of phytoremediation techniques

1: Are the contaminants within reach of the plants?

The roots of the plants need to be within reach of the contamination in order to take it up. If the plants cannot take up the contamination nothing but prevention of soil corrosion and hydraulic control is obtained. Root depths vary with plant species, why appropriate species has to be selected due to the contamination depth.

2: Are plants able to take up the contamination?

The uptake by plants occurs for most substances with the transpiration water. However, only non-adsorbing (neutral, polar) compounds are also translocated upwards, out of soil and groundwater. This transport can easily be calculated (Trapp 2007).

3: Is the contamination volatile?

This refers to box 4b of flow chart A. The data obtained should be sufficient to answer this question.

4: Can the contamination be degraded in soil or plants or in the rhizosphere?

This refers to box 4a of flow chart A. The data obtained should be sufficient to answer this question.

Risk assessment of given application:

Some contaminants may degrade into products which are even more problematic than the mother compound. An example are ferro- and ferricyanides, which decompose to highly toxic free cyanide. Before implementing phytoremediation, the different degradation pathways and metabolites have to be assessed. Further, a risk assessment should be carried out for the application of phytoremediation. Issues like playing kids, collection of fruits and grassing wild life should be considered.

5. General considerations

Besides location, depth and extent (concentration) of the contamination, properties of the pollutants directly affect the probable success of phytoremediation projects. As can be seen from the flow chart, toxicity to plants, degradation rates and volatility are compound properties that affect type and success of phytoremediation. While volatility directly depends on the partition coefficient air-to-soil or air-to-leaf and can be estimated accurately from the physico-chemical properties of the compound, namely partition coefficient octanol-water and air-water (which in turn can be estimated from water solubility and vapour pressure of the compound). Such properties can be looked up easily in handbooks (Lyman et al. 1990) or estimated by structure-property estimation routines (ACD 2012). More difficult to get are data on phytotoxicity and degradation. These have to be tested in laboratory and furthermore also depend on the conditions at site (for example, water uptake, temperature, availability of electron acceptors such as oxygen). To guide the user of this report, we collected data on phytotoxicity from earlier studies and papers. Much of these data has not been published except in research project reports.

5.1 Phytotoxicity

The following data have been collected in and from several projects, many of them unpublished student projects. The tests are obtained by the willow tree transpiration acute toxicity test (Trapp et al. 2000). Trees are grown in a 500 mL Erlenmeyer flask and exposed to the toxin in solution. That is a kind of worst-case approach. Thus, if trees survive the conditions in this test, it may safely be assumed that they can grow outdoors on soil or groundwater polluted with a similar concentration. These measured toxicity data on trees and other plants can be used to judge the feasibility of phytoremediation projects with respect to phytotoxicity at site. As can be seen, trees are generally rather insensitive organisms and can stand quite high doses of soil- and groundwater pollutants. This is of certainly an advantage for phytoremediation projects. And in fact, there are few sites known where trees or forests died due to soil- or groundwater pollution. Contrary, most investigated sites have a wild population of pioneer trees (e.g., birch or willows), a feature that is used for phytoscreening by tree coring (cf. Deliverable 4.2).

5.2 Data collection

The presented tables are extracts of the ModelPROBE Deliverable 4.4. We cite from there. New data was added as available.

Table 5.1a Toxicity (estimated EC50) of the frequent groundwater pollutants PCE/TCE/DCE, MTBE, BTEX tested in the willow tree toxicity test. EC50 given in mg/L hydroponic solution, or mg/L irrigation water (sand), or mg/kg (soil). Endpoint: inhibition of transpiration.

Compound	Species	Medium	EC50	Conditions	Reference
TCE	<i>hybrid poplar</i>	nutrient solution	131	14 d	Dietz & Schnoor 2001
TCE	<i>S. viminalis</i>	nutrient solution	100	96 h	Clausen et al. 2006
PCE	<i>hybrid poplar</i>	nutrient solution	38	14 d	Dietz & Schnoor 2001
PCE	<i>S. viminalis</i>	nutrient solution	130	96 h	Clausen et al. 2006
c-DCE	<i>S. viminalis</i>	nutrient solution	107	96 h	Aelion 2006
TCE	<i>S. viminalis</i>	nutrient solution	55	96 h	Aelion 2006
PCE	<i>S. viminalis</i>	nutrient solution	130	72 h	Aelion 2006
PCE + TCE + c-DCE mix	<i>S. viminalis</i>	nutrient solution	10	168 h	Aelion 2006
MTBE Methyl- <i>t</i> -butylether	<i>S. viminalis</i>	nutrient solution	approx. 1000	72 h	Trapp & Karlson 2001
MTBE	<i>S. viminalis</i>	nutrient solution	1740	88 h	Eva Seeger, unpublished
Chlorobenzene	<i>S. viminalis</i>	nutrient solution	210 and 114	88 h	Eva Seeger, unpublished
Nitrobenzene	<i>Populus trichocarpa</i>	nutrient solution	205	72 h	Johannsson 2000
Benzene	<i>S. viminalis</i>	nutrient solution	292	88 h	Eva Seeger, unpublished
Toluene	<i>S. viminalis</i>	nutrient solution	< 500	216 h	Kolmorgen 2002

Table 5.1b Toxicity (estimated EC50) of frequent soil pollutants (petroleum products, PAH and TBT) tested in the willow tree toxicity test. EC50 given in mg/kg (soil). Endpoint: inhibition of transpiration.

Compound	Species	Medium	EC50	Conditions	Reference
Diesel (sum of hydrocarbons)	<i>S. viminalis</i> <i>x</i> <i>schwerinii</i>	contaminated soil	3910 soil	144 h	Trapp et al. 2001
Gasoline ROZ 95 (sum hydrocarbons)	<i>S. viminalis</i> <i>x</i> <i>schwerinii</i>	spiked soil	< 1000 soil	96 h	Trapp et al. 2001
Naphthalene	<i>S. alba</i>	nutrient solution	32.5 (solubility)	134 h	Thygesen & Trapp 2002
Phenanthrene	<i>S. alba</i>	nutrient solution	>1.66 >solubility	300 h	Thygesen & Trapp 2002
Benzo(a)pyrene	<i>S. alba</i>	nutrient solution	>> 0.05 > solubility	300 h	Thygesen & Trapp 2002
PAH (mix)	<i>S. viminalis</i> <i>x</i> <i>schwerinii</i>	contaminated soil	> 1000	300 h	Thygesen & Trapp 2002
TBT-Cl	<i>S. viminalis</i>	nutrient solution	0.1-1 > 1	pH 4 72- 288h pH 7 72-288 h	Trapp, Ciucani, Sismilich 2004
TBT-H	<i>S. viminalis</i>	nutrient solution	1-10	pH 4 and 7 72- 288h	Trapp, Ciucani, Sismilich 2004
TBT	field crops	contaminated sediments	> 33	3 months (field test)	Novak & Trapp 2005

Table 5.1c Toxicity of phenolic compounds tested in the willow tree toxicity test. EC50 given in mg/L hydroponic solution, or mg/L irrigation water (sand), or mg/kg (soil). Endpoint: inhibition of transpiration.

Compound	Species	Medium	EC 50	Conditions	Reference
Phenol	<i>Salix viminalis</i>	nutrient solution	600 400	72 h 120 h	Ucisik & Trapp 2006
Phenol	<i>S. viminalis</i>	sand	800 600	72 h 120 h	Ucisik & Trapp 2006
4-chlorophenol	<i>S. viminalis</i>	nutrient solution	32.2	72 h	Ucisik & Trapp 2008
4-chlorophenol	<i>Populus trichocarpa</i>	nutrient solution	84	72 h	Johannsson 2000
3,5-dichlorophenol	<i>Populus trichocarpa</i>	nutrient solution	16.4	72 h	Johannsson 2000
3,5-dichlorophenol	<i>Populus trichocarpa</i>	nutrient solution	16.4	72 h	Johannsson 2000
2,4-dichlorophenol	<i>S. viminalis</i>	nutrient solution	10	72 h	Ucisik et al 2007
2,4-dichlorophenol	<i>S. viminalis</i>	nutrient solution	5.8-7.8 6.5	72 h 163 h	Trapp et al. 2000

Table 5.1d Toxicity of heavy metals tested in the willow tree toxicity test. EC50 given in mg/L hydroponic solution, or mg/L irrigation water (sand), or mg/kg (soil). Endpoint: inhibition of transpiration.

Heavy metals	Species	Medium	EC50	Conditions	Reference
As (arsenate)	<i>S. viminalis</i>	solution	3	72 h	unpublished, student work
Copper (Cu ²⁺)	<i>S. viminalis</i>	solution	0.9	72 h	Boeck 2004
Copper (Cu ²⁺)	<i>S. viminalis</i>	solution	0.4 to 1	72 h to 240 h	Wibroe & Staal-Thomsen 2006
Cadmium (Cd ²⁺)	<i>S. viminalis</i>	solution	1.5	240 h	Wibroe & Staal-Thomsen 2006
Cd + Cu mix	<i>S. viminalis</i>	solution	1 and 1	240 h	Wibroe & Staal-Thomsen 2006
Selenate, selenite	<i>Salix</i> sp.	nutrient solution	> 0.85	168 h	Yu & Gu 2008

Table 5.1e Toxicity of other inorganic compounds tested in the willow tree toxicity test. EC50 given in mg/L hydroponic solution, or mg/L irrigation water (sand), or mg/kg (soil). Endpoint: inhibition of transpiration.

Compound	Species	Medium	EC50	Conditions	Reference
Inorganic compounds					
NaCl salt	<i>S. viminalis</i>	nutrient solution	1000 600	72 h 240 h	Trapp et al. 2008
Nano-TiO ₂	<i>S. viminalis</i>	solution	> 100	210	Seeger et al. 2009
nano-zero-valent iron	<i>Populus deltoids</i> × <i>nigra</i>	solution	> 200 < 1000	28 days	Ma et al. 2013
Cyanides					
KCN (HCN)	<i>S. viminalis</i>	nutrient solution	2 (CN) < 2	72 h 190 h	Larsen et al. 2004
KCN (HCN)	<i>S. viminalis</i>	sand	20 (CN)	72 h	Larsen et al. 2005
KCN (HCN)	<i>S. babylonica</i>	nutrient solution	10 (CN) 6.5	96 h 192 h	Yu et al. 2005
KCN (HCN)	<i>S. babylonica</i>	sand	18.6	216 h	Yu et al. 2005
KCN (HCN)	<i>Sorhum bicolor</i>	sand	<< 50 (CN)	360 h	Trapp et al. 2003b
Ferrocyanide, ferricyanide	<i>S. viminalis</i>	solution	> 10	> 1200 h	Larsen & Trapp 2006
Prussian blue (iron CN; ferri ferrocyanoide)	<i>Populus tricarpa</i>	nutrient solution	2500 1000	72 h 290 h	Trapp, Koch, Christiansen 2001; Trapp & Christiansen 2003

6. Degradation

Degradation by plant cells has been tested in a closed-bottle standard test (see, e.g., Trapp et al. 2003, Yu et al. 2004, Larsen et al. 2004). Results are listed below.

Overall, the results obtained with this closed-bottle degradation test were disappointing. Plants are different from bacteria. Most bacteria are heterotrophic, and due to the rapid evolution of microbial enzymes, a wide variety of degrader genes has been determined. Plants do not live and grow on xenobiotic compounds and, if at all, have enzymes for detoxification (Trapp et al. 2007 in Heipieper HJ). Their capacity for degradation is thus generally small. For example, none of the investigated plant species (28 species from a couple of genera) was able to degrade MTBE (Trapp et al. 2003). One substance, however, that perhaps all plants can degrade, is hydrogen cyanide (HCN) (Yu et al. 2004, Larsen et al. 2004).

Another list with results from the plant cell degradation test is found in Trapp and McFarlane (1995), presented by H. Sandermann. Those data might be affected by bacterial contamination (personal communication).

Indeed, it may well be that degradation by plants was intended to be measured in pot or field experiments, but in reality, the enzymes belonged to microbes living on or even inside plant material. In fact, several hundred different microbial species could be identified as endophytic (ENDEGRADE project report, Karlson et al. 2004). Consequently, the technique to “infect” plants with degrader strains has been developed and used for remediation (Barac et al. 2004, Van der Lelie et al. 2001, 2002).

6.1 Root zone degradation

Cited from Trapp and Karlson (2001): The root zone (rhizosphere) is a very lively part of the world, with fungi, roots and bacteria acting together. Within a distance < 1 mm to roots live approx. 1.2×10^{11} cells per cm^3 , in 2 cm distance only 1.3×10^{10} (Paul and Clark, 1989). About 5 to 10% of the roots surface is covered with bacteria. Roots live in symbiosis with fungal mycorrhiza. Their mycelium is again covered with bacteria (Romantschuk et al., 2000). Growing roots may transport inoculated bacteria through soil (Karlson et al., 1995). Besides forming a habitat for microorganisms, plant roots also provide nutrients, e.g., sugars, in exchange for phosphate (fungi) or nitrogen (N_2 -fixation). Mulberries *Morus rubra* L. growing at PCB-polluted sites, excrete considerable amounts of phenolic compounds, which probably support the growth of PCB-degrading bacteria (Fletcher and Hedge, 1995).

Four years after the Gulf war and the oil spill in Kuwait, wild flowers (mainly Compositae, e.g., *Senecio glaucus*) were growing in sand polluted with up to 10% petroleum. Roots of these plants were associated with millions of oil degrading bacteria (*Arthrobacter*), which took up and detoxified alkanes and aromatic hydrocarbons. The roots practically were free from oil (Radwan et al., 1995).

Trichloroethylene, TCE, can be degraded bacterially by co-metabolic oxidation. Nonetheless, it is rather persistent in the environment. Under anaerobic conditions, reductive dehalogenation is accompanied by the formation of the carcinogenic metabolite vinyl chloride. TCE degradation has been shown for several plant species, among them poplars

(Newman et al., 1997). In laboratory experiments with 35 cm tall poplar cuttings, fast uptake was observed, but the main fate process was volatilization from leaves (Burken and Schnoor, 1998). However, in field-scale experiments with three-year old poplars of 6 m height, the main fraction of TCE was mineralized in the root zone. Up to 99% of TCE added with artificially contaminated groundwater were removed, and less than 9% escaped to atmosphere (Newman et al., 1999).

We are currently repeating these experiments in a controlled laboratory study with trees depleted of any chlorid. The occurrence of chloride can thus be used as tracer for aerobic TCE degradation. It must be noted that so far we neither found an increased loss of TCE by trees, nor a measurable formation of chloride in the vicinity of trees. Thus, we are somewhat sceptic towards the results obtained by Newman et al. (1997). The experiments are ongoing within the frame of the Timbre project, and we hope we can report the final results later 2014.

6.2 Indirect stimulation of degradation by aeration

The average rooting depth of trees is 1-2 m, which is much less than usually expected, and almost 90% of tree roots are found in < 0.6m depth. Poor aeration will lead to a smaller root density. Poplars root to a depth between 0.8 and 2.43 m (Dobson and Moffat, 1995). Although willows survive permanent flooding and water saturated soils, the roots will not grow deep into the groundwater (Larcher, 1995). Thus, degradation of chemicals in groundwater or deeper soil by plant cells is not expected to be of high relevance. But nonetheless, plants can by their activities indirectly stimulate degradation in root zone and also deeper layers:

Evapotranspiration of water means less water-filled pores, more gas-filled pores and a much faster flux of gases through soil (oxygen diffusion in gas-filled pores is about 300000 times faster than in water filled pores). Thus, simply by transpiring soil water and thus producing air-filled pores, plants can increase the electron acceptors needed for the degradation of many pollutants, a prominent example being petroleum products such as gasoline or jet fuel (Wiedemeier et al. 1999).

Additionally, some wetland plants, among them basket willow (*Salix viminalis*), possess a ventilation system for roots (Grosse et al., 1992, 1996). For *Phragmites*, oxygen fluxes of up to $8 \text{ g m}^{-2} \text{ d}^{-1}$ have been determined (Brix et al., 1996). For basket willow, fluxes are around $0.5 \text{ g m}^{-2} \text{ d}^{-1}$ (own calculation) and therefore only relevant in water-saturated soils.

We will consider aeration processes by plants in detail in the specific considerations for the Timbre sites.

6.3 Data collection

The following Table 6.1 lists the experience of PhD- or student projects at DTU made with the closed-bottle degradation test for plants.

Table 6.1 Results from the closed-bottle degradation test for plants made at DTU.

Degradation by plants	FeCN (iron cyanide, both FeII and FeIII-species)	Larsen and Trapp 2005
Degradation by plants and bacteria	HCN, chlorobenzoic acid, chlorobenzene	Larsen et al. 2004, Yu et al. 2004, Deavers et al. 2010
Degradation by bacteria only	Benzene, toluene, xylene, phenol, TBT, gasoline and petroleum, polycyclic aromatic hydrocarbons, detergents, chlorophenol, trichloroethene	Ucisik et al. 2006, 2007, 2008 and others
Recalcitrant or persistent	MTBE	Trapp et al. 2003

References do not cover all results.

7. Mass balances and modeling

The long-term fate of chemicals residing in soil (Hunedoara) or groundwater (Szprotawa) can be estimated with mathematical models that rebuild the transport and loss processes. These mass balances can also consider phytoremediation as treatment option and thus give an estimate of the feasibility, time scale and success.

Typical loss processes for soil and groundwater pollutants are leaching from top soil, degradation (including biodegradation), and uptake into plants. Figure 7.1 gives a general overview of the mass balance of chemicals in soil with trees.

We had, as task in WP 4, prepared and distributed Timbre milestone “Mathematical models to quantify biodegradation, after identification of rate-limiting processes”. The model was adapted to the conditions at the Timbre sites. This means both that site-specific input parameters were chosen, but also that only the relevant processes at site were considered, and that is: Plant uptake of heavy metals from soil at Hunedoara, and plant uptake of alkanes (as model compound for jet fuel) from groundwater plus biodegradation in groundwater for Szprotawa. We also simplified the models from dynamic versions to time-constant versions (i.e. no change of parameters with season, instead average annual data as input). Seen over long periods (decades to centuries) a detailed consideration of time (daily or weekly data input) is resource demanding and not relevant for the results.

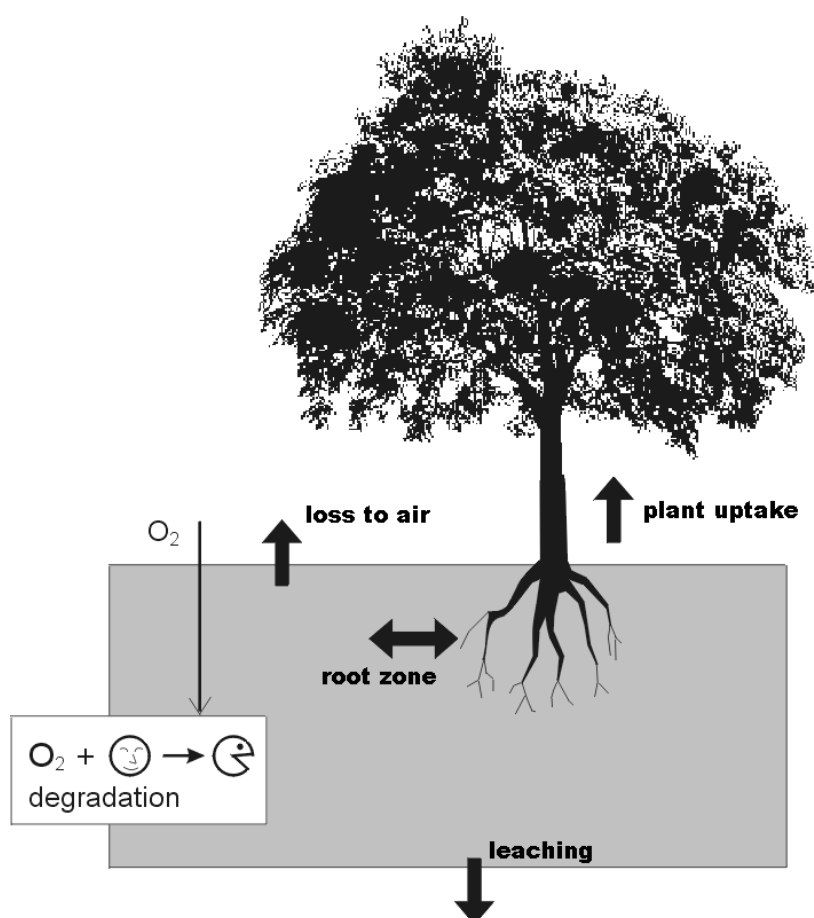


Figure 7.1 Process overview for the mass balance of chemicals in soil with trees.

7.1 Phytotoxicity

Soil compartment

The central compartment is the soil, which consists of a well-mixed one meter thick layer. All calculations are made for 1 m². Initially, a completely mixed and homogeneously contaminated layer of various, given depth is assumed. For removal from groundwater, an aquifer of measured thickness was taken.

Sorption

The presence of the compounds in GW or solution is a pre-requisite for metabolism, plant uptake and leaching. The fraction present in GW or solution is expressed as K_d -value (L water per kg soil or aquifer material) and was calculated from measured concentrations

$$K_d = \frac{\text{total concentration in bulk soil } C_B}{\text{concentration in solution } C_S} \quad (\text{measured})$$

Then, for any concentration in soil, concentration in solution C_S (mg/L) can be derived from the total bulk soil concentration C_B (mg/kg)

$$C_S = C_B / K_d$$

Leaching

Leaching to groundwater can be calculated from the fraction of chemical solved in soil solution times the specific flow downwards. The water flow that infiltrates follows from the water balance:

$$\text{Infiltration} = \text{precipitation} - \text{evapotranspiration} - (\text{runoff}) - (\text{storage}) = P - Q$$

Storage and runoff are set to zero. The infiltration is then critically influenced by the evapotranspiration.

The loss of compound from the soil compartment to deeper soil layers is then

$$dm/dt = - (P - Q) C_S$$

where m is the mass of chemical in soil (kg), t is the time and Q is the water flow through the xylem. In our calculations for Hunedoara, we neglected leaching because it is not a remediation but a transfer process. In Szprotawa, chemicals reside in GW, and leaching from top soil is thus not a relevant process.

Uptake into plants

For Hunedoara, we could use measured concentrations of heavy metals in wood to quantify plant uptake, but for Szprotawa we calculated uptake this process. Uptake into plants and transport out of the soil is with the transpired water. Hereof, loss from GW is only with water taken up from groundwater. We do not know the exact amount of water that trees at Szprotawa take from GW. It is higher in dry summers, and with high groundwater table. A default value of 100 L/m² for water uptake from groundwater was chosen for Szprotawa.

Generally,

$$dm/dt = Q \times C_s$$

where m is chemical mass (mg), Q is water flow (L/d) and CS is concentration in soil solution or GW (mg/L).

Chemicals taken up into plants can be accumulated, metabolised, or gas out into atmosphere, and a detailed model for trees has been developed (Trapp 2007). Even if degradation is slow, volatilisation from aerial plant parts is a very important fate process for the volatile components of jet fuel present in Szprotawa. Experiments and model calculations show that volatile compounds, like light alkanes from jet fuels, do not accumulate in trees but are quickly lost from branches and trees (Trapp 2007, Baduru et al. 2008). We do thus not expect that an accumulation of the pollutants present in the GW of Szprotawa in trees occurs. Calculations are not shown here but were done with the “Fruit Tree Model” (Trapp 2007). We do also not expect that accumulation in food crops would be problematic (WP 6 Timbre), due to the high volatility of the contaminants.

Concentrations in wood were measured, see Timbre deliverable 4.2. In the first sampling campaign at Szprotawa, samples from pines and birches were collected and only toluene could be detected in these tree cores. In the second sampling campaign, aspens and willows were sampled and in addition analyzed for MTBE and TCE. Toluene was detected in all samples, ethyl benzene, m, p, o-xylene were detected in most of the samples (26 and 24 out of 35 samples, respectively) and benzene and naphthalene in some of the samples (eight out of 35 samples). Only in few samples from Hunedoara, BTEXN could be detected (Algreen et al. 2013, Timbre deliverable 4.2).

7.2 Degradation in soil

Short- and long-chain hydrocarbons (alkanes) can be utilised by a large number of bacteria. The breakdown is initiated by oxidation of the terminal carbon. Molecular oxygen is required for the breakdown (Schlegel 1986). It has been shown that alkanes and BTEX are also degraded in the absence of oxygen, but under presence of nitrate and sulphate. Under certain conditions, even methanogenic degradation may occur (Wiedemeier et al. 1999).

The spill in Szprotawa is more than ten years old. Nitrates, reducible irons and sulphate have therefore very likely already been used by bacteria and are no more available as electron acceptors. Degradation is then limited by the downwards transport and availability of new electron acceptors - oxygen from air.

Immediate reaction model for the degradation of hydrocarbons

When electron acceptors limit the break-down of compounds, the "immediate reaction model" may be used to calculate degradation (Wiedemeier et al. 1999). It is based on the following theoretical considerations:

- Reaction rates of bacterial enzymes are fast compared to the transport rates of oxygen
- The substrate (BTEX, alkanes) is present in excess
- The biomass is in steady state
- The degradation is limited by oxygen transport.

All oxygen arriving at the spill is used for oxidation, for octane (C_8H_{18}) as example compound:



Octane has a molar weight of approx. 114 g/mol. A complete oxidation of one mole requires 25 moles or 400 grams oxygen. With other words, 3.5 g O_2 are needed to oxidise 1 g octane.

Bacteria prefer BTEX and low-molecular-weight alkanes over long-chain alkanes (Wiedemeier et al. 1999). This means that the long-chain alkanes will - relatively to the other pollutants - accumulate in the soil. This bacterial preference is not considered in the model, all carbohydrates are given the same zero-order degradation rate.

Influence of trees on biodegradation

We did the calculations for willow trees, for which we have all necessary data determined within this and other projects. One expected benefit from willow trees is the facilitated transport of oxygen to the root zone. Basket willow trees (*Salix viminalis*) are known to grow under conditions where they are periodically flooded (Ellenberg 1979). The reason is that willows are adapted to oxygen deficiency in soil by a ventilating tissue (aerenchym), which has a large gas-filled intercellular space (up to 60%). These pores allow oxygen to diffuse downwards. Oxygen supply to roots by gas diffusion is only effective over short distances. Willows have therefore established a pressure driven ventilation system. For *Salix viminalis*, a surplus flux to the root zone of 2.1 to 4.3 $\mu\text{mol } O_2$ per 40 cm-cutting and hour was measured (Grosse et al. 1996).

Another positive property of willows is that they will transpire water. Therefore, the groundwater table sinks, and parts of the polluted soil layer become aerated. This is not

explicitly considered in the calculations for Szprotawa, because this requires a complete 3D-model for groundwater flow.

Transport of oxygen to groundwater

Calculations were made for unit area $A = 1 \text{ m}^2$. The unit flux J ($\text{kg d}^{-1} \text{ m}^{-2}$) of oxygen is calculated with Fick's 1st Law of diffusion:

$$J = A D/x \Delta C$$

ΔC is the concentration gradient between soil surface and 1 m depth, where $C(\text{O}_2) = 0$. D is the diffusion coefficient (in liquid or gas, see below). For gas phase diffusion, the gradient ΔC is then 20.95 vol. % O_2 per m^3 air or 300 g/m^3 air. For the diffusion in water, ΔC is the saturation concentration (10.0 mg/L at 15 °C).

Three scenarios were assumed, namely 1) worst case = water saturated, no trees; 2) water-saturated but with trees and 3) aerated soil with trees.

Scenario 1: Worst case: water saturated soil, no vegetation

The diffusion coefficient of molecular oxygen D_w in water at standard conditions is $1.728 \times 10^{-4} \text{ m}^2 \text{ d}^{-1}$ (Trapp and Matthies 1998). The effective diffusion coefficient in soil D_{eff} is calculated by

$$D_{\text{eff}} = D_w * P_w * T = 1.728 * 10^{-4} \text{ m}^2 \text{ d}^{-1} * 0.5 * 0.12 = 10.4 \times 10^{-6} \text{ m}^2/\text{d}$$

where P_w is the volume fraction of water-filled pores (50%) and T is a labyrinth or tortuosity factor (Jury et al. 1983). The flux of oxygen over a distance of one meter through the surface of one square meter is 36.5 mg per year. This amount is just enough to oxidise 10 mg octane per year and square meter - or less than $7 \text{ } \mu\text{g}$ per year and kg soil (if the soil has a density of 1500 kg/m^3 and 1m soil layer is polluted). Thus, biodegradation in water-saturated areas without vegetation is negligible.

Scenario 2: Water saturated soils, additional oxygen surplus by willow trees

In this scenario, the oxygen surplus from willow roots is considered. As basis for a calculation, 3 $\mu\text{mole O}_2$ for a 20 g willow cutting and per hour are taken. In the field, a mass of 8 kg willows per m^2 is what can be achieved (Perttu and Kowalik 1997), and flux of oxygen is then 400 times higher. The unit flux per year ($\frac{1}{2}$ time) is then $168.2 \text{ g m}^{-2} \text{ a}^{-1}$. This is sufficient to oxidise 48 g octane per square meter and year.

Scenario 3: Aerated soil

In this scenario, it is assumed that the pollutants reside in the aerated soil zone. The effective diffusion of gases in soil D_{eff} can be estimated from the diffusion in air D_A (Trapp and Matthies 1998):

$$D_{\text{eff}} = D_A * P_A * T = 1.65 \text{ m}^2/\text{d} * 0.2 * 0.12 = 0.04 \text{ m}^2/\text{d}$$

Measured values of D_{eff} for 25% air pores are up to $0.1 \text{ m}^2/\text{d}$ (Schachtschabel et al. 1984).

The oxygen flux is then

$$J = A D/x \Delta C = 1 \text{ m}^2 \times 0.04 \text{ m}^2 \text{ d}^{-1} / 1 \text{ m} \times 300 \text{ g/m}^3 = 12 \text{ g/day or } 4380 \text{ g/year}$$

The flux is 120 000 times larger than in water-saturated soil. 4380 g oxygen can oxidise 1217 g octane per year.

Additionally, every liter of water that is transpired is replaced by soil gas stemming from air. A transpiration of 400 L water per m^2 and year would thus deliver an additional 84 L (119 g) of oxygen downwards. This advection process is small compared to diffusion, but still more relevant than the transport in aerenchyma (the transport of air downwards in aerenchyma is partly driven by this replacement of water with air).

The calculated value for oxygen diffusion is in excellent agreement with the measured oxygen transport and aquifer reaeration by Neale et al. (2000). These authors give a reaeration flux ranging from 11 to 12 g oxygen per m^2 and d and for conditions typical for natural environments (Neale et al. 2000). The authors also used a similar gas flux model as we have used here, and come to a similar result.

This rough calculation can also be compared to measured fluxes in Schachtschabel et al. (1984). For a depth of 80 cm, the maximum oxygen diffusion rate was $2.88 \text{ g d}^{-1} \text{ m}^{-2}$ or 1051.2 g/year. The average is below $0.72 \text{ g d}^{-1} \text{ m}^{-2}$ or 262 g/year. Probably, in this setting a large fraction of the oxygen was consumed before it reached 80 cm depth.

A comparison of the scenarios shows that the most important effect of trees for aeration of GW is the transpiration of water and thus aeration of soil. If no gas pores are available, transport of oxygen is negligible. The transport of air in the aerenchyma of plants such as willows is comparatively small.

To allow for some oxidation of soil material underway (humic compounds, root exudates, decaying roots) we set the oxygen flux in the calculations for Szprotawa to GW to $10 \text{ g/m}^2/\text{d}$.

8. Evaluation of phytoremediation at the Hunedoara site

The principles, methods, data and models described in the previous chapters can be used to judge the feasibility of phytoremediation at the Timbre site Hunedoara in Romania.

8.1 Flow chart Hunedoara

The flow chart shown in section 4.1 is applied for the situation in Hunedoara

Step 1: Do usage of the area prohibit plant growth?

The future land use of the areas is nothing decided in WP 4. But parts of the area could be used to establish a forest or park area, and other parts could be used for residential areas or agriculture, thus, principally the answer is no.

Step 2: Is time of essence?

For areas where a park or forest shall be established, no.

Step 3: Are the conditions at site toxic to plants?

There are several ways to answer this question.

a) By inspection of plant growth at site.

This was done within the Timbre project. Available is “A9. Information/data on nutrient availability for plants (identification of different soil types including chemical soil parameters, i.e. pH-value, content of lime, water and plant nutrients N, P, K)” as well as “General information on soil type, GIS soil maps already available (UTCB), more precise data to come during the project through field analyses (DTU)”.

In order to retrieve this information, the presence of vegetal species was determined during the first field trip. The species were identified using Rothmaler's Exkursionsflora (Jäger et al. 1988) and the BLV plant guide (Schauer and Caspari 1978) and, in (rare) case of doubt, google pictures. The presence of species was processed for a geoecological analysis by determining the indicator values of these plants. This is a well-established method in plant ecology and geoecology. The list of Ellenberg (1979) was used.

A large number of plant species was identified and allows a characterisation of the area: Summarized, the classification of the site is: Warm, good water supply, neutral to slightly alkaline soil pH and good supply with nitrogen. This makes the soil and conditions at site favourable for agricultural use. This holds of course only for the areas where the soil is not covered with anthropogenic ruins. Also, trees were sampled during the campaigns for WP 4 (results see Algreen et al., deliverable 4.2). There were no obvious signs of toxicity.

Summarized, by visual inspection of the site, there were no indications of intoxicification of plants. However, this is of course no proof for its absence: if plant growth is inhibited by contaminants, then the presence of plants cannot be used to indicate this because plants would probably not grow at all.

Therefore, we used a second method.

b) By comparison of measured concentrations of contaminants in soil with the known toxicity data presented in section 5.1.

Chemicals in soil were analysed by various means. The results of the analysis was collected within the Timbre project (Timbre 2013a, available as excel file "HUNEDOARA_mastersheet.xls"). Most relevant for Hunedoara are some of the heavy metals, but also measured concentrations of BTEX and PAH are shown (maximum, mean, median). Table 8.1 shows the results of the chemical analysis. RXF-data are also available, and are often higher (Ni, Mn, Zn).

Relevant for a judgement of phytotoxicity is the dissolved concentration in soil pore water, because this refers to the solution concentrations in the phytotox tests section 5.1. To get the same basis, the concentrations in soil were recalculated using the Kd. The Kd (defined above) of heavy metals depends critically on pH, and also on other soil parameters, such as organic carbon content.

Values of pH at Hunedoara were provided in the file "Soil_conc_Hunedoarareport.pdf". The pH-values given there range from 7.65 to 8.97 (Table 2, 2007), with a median pH of 8.22. The same source also contains measurements of organic matter ("humus"), values range between 0.42% and 3.49%, with median at 1.11%. These data were used to calculate Kd-values with the linear regressions of Sauvé at al. (2000), Table 8.2. For elements not covered by Sauvé's regressions, we used the pH-specific Kd-values given in the US-EPA 2012 Soil Screening Guidance, that is: Arsenic As at pH 8 Kd is 31 L/Kg; chromium III (Cr³⁺) 4 300 000 L/kg. No data was found for manganese Mn.

The Kd of organic compounds is usually calculated from the Koc (partition coefficient organic carbon to water, Trapp and Matthies 1998), which in turn can be estimated from the log Kow. The European Commission (EC 1996, 2003) recommends the regression

$$\log Koc = 0.81 \times \log Kow + 0.1$$

which is used here. Moreover,

$$Kd = orgC \times Koc$$

orgC is the organic carbon content (here in the unit g/g) which is related to soil organic matter OM by

$$orgC = 1.722 OM \quad (\text{Lyman et al. 1990}).$$

Values for log Kow (taken from Trapp and Matthies 1998, which in turn cite Rippen 1990) and calculated values for Koc and Kd (using the median OM from Hunedoara, then orgC = 0.0065 g/g) are shown in Table 8.3.

The concentrations in soil pore water result by dividing the concentration in bulk soil (mg/kg dw) with the Kd. And these values were then compared in Table 8.4 with the measured toxicity data for trees. If toxicity-tests with contaminants in soil were available, these were used without recalculation. This was the case for petroleum (where the toxicity tests with trees in soil were available from a former tank station, with gasoline and diesel in soil, and from controlled studies in laboratory (Trapp et al. 2001).

The Table 8.4 shows that the peak (maximum) concentrations at Hunedoara are far above what is known as toxic, and this holds both for arsenic, and for gasoline. The median concentrations would not exceed the toxic threshold levels. Toxicity of heavy metals (Cd, Cr, Cu, Ni, Pb, Zn) would, as far as it can be judged from the few available toxicity data, not be problematic for the growth of trees. This is due to the very heavy binding (high Kd) of the heavy metals in Hunedoara soils.

Table 8.1 Measured concentrations (chemical analysis) in top soil of Hunedoara, unit mg/kg.

	Maximum	Mean	Median
As	4281	222	40
Cd	20	5.2	2.8
Cr	187	82	65
Cu	1255	236	144
Pb	2982	583	229
Ni	66	47	46
Mn	23881	7049	5807
Zn	1599	556	477
TPH-GC *	175700	7897	237
Naphthalene	2533.0	58.6	0.5
Fluoranthene	146.0	11.9	1.0
Benzo(a)pyrene	45.8	7.4	1.4
Total PAH	92137.0	2437.6	13.6
Benzene	3.4	0.22	0.013
Toluene	2.9	0.21	0.022
Xylene	2	0.2	0.03
Sum BTEX	7.1	0.59	0.055

* total petroleum hydrocarbons analyzed by gas chromatograph

Table 8.2 Distribution coefficient Kd between soil matrix and soil pore water (L/kg) calculated from pH and soil organic matter (OM, in %) with the log-linear regressions of Sauvé et al. (2000).

Kd Sauvé L/kg	pH	OM (%)	log OM	a	b	constant	log Kd	Kd
Cd	8.22	1.11	0.045	0.48	0.82	-0.65	3.33	2151.6
Cu	8.22	1.11	0.045	0.21	0.51	1.75	3.50	3157.3
Ni	8.22	1.11	0.045	1.02	0.8	-4.16	4.26	18224.6
Pb	8.22	1.11	0.045	0.37	0.44	1.19	4.25	17837.8
Zn	8.22	1.11	0.045	0.6	0.21	-1.34	3.60	3995.0

Table 8.3 Literature data for log Kow (Trapp and Matthies 1998), calculated Koc and Kd values (orgC = 0.00645 g/g, median Hunedoara).

	log Kow	log Koc	Koc	Kd
Benzene	2.12	1.82	66	0.42
Toluene	2.66	2.25	180	1.16
Xylene	3.18	2.68	474	3.1
Naphthalene	3.37	2.83	676	4.4
Fluoranthene	5.13	4.26	18001	116.0
Benzo(a)pyrene	6.04	4.99	98265	633.4
average BTEX				1.55
average PAH				251.3

The concentrations in soil pore water result by dividing the concentration in bulk soil (mg/kg dw) with the Kd. These values were then compared in Table 8.4 with the measured toxicity data for trees. If toxicity tests with contaminants in soil were available, these were used without recalculation.

Table 8.4 Maximum and median concentrations in Hunedoara soil (mg/kg); Kd-values (L/kg) from Table B and C; calculated concentration in soil solution (mg/L); toxicity data for trees (EC50, mg/L) from section 5.1; and judgement of toxicity (if concentration above EC50).

	Maximum soil mg/kg	Median soil mg/kg	Kd L/kg	Maximum solution mg/L	Median sol. mg/L	EC50 mg/L	Judgement
As	4281	40	31	138.1	1.30	3	toxic
Cd	20	2.8	2152	0.7	0.001	1	no
Cr	187	65	4300000	6.0	0.000	nd	nd
Cu	1255	144	3157	40.5	0.045	1	no
Pb	2982	229	17838	96.2	0.013	nd	nd
Ni	66	46	18225	2.1	0.003	nd	nd
Zn	1599	477	3995	51.6	0.119	nd	nd
TPH-GC	175700	237				3910 (soil)	toxic
Naphthalene	2533	0.5	4.4	81.7	0.119	32.5	toxic
Fluoranthene	146	1.0	116	4.7	0.008	nd	
Benzo(a)-pyrene	46	1.4	633	1.5	0.002	0.1	
Total PAH	92137	13.6				1000 (soil)	toxic
Benzene	3.4	0.013	0.42	0.11	0.031	292	no
Toluene	2.9	0.022	1.16	0.09	0.019	500	no
Xylene	2.0	0.030	3.1	0.06	0.010	nd	no
Sum BTEX	7.1	0.055	1.55	0.23	0.035	< 921 (soil)	no

Thus, we can finally answer the question step3

Step 3: Are the conditions at site toxic to plants?

Yes, at the “hot spots”.

If the site is cleaned up, i.e. the hot spots removed, or if only the less contaminated area is planted, then phytoremediation is still possible.

Step 4. Degradation rates

BTEX, petroleum and PAH can be (at least partly) degraded. This will be handled at the Szprotawa case. Heavy metals, which are a major problem at the site, are not degradable.

Step 5. Volatilization

BTEX, petroleum and the lower weight PAH are known as volatile. This can be calculated by, e.g., the model of Jury et al. (1987). Heavy metals, which are a major problem at the site, are ionic and do not volatilize (except Hg and Se).

Step 6. Mass balance model

The mass balance model will be done for the heavy metals at site, because these are the longest lasting form of pollution. It will be done for a phytoremediation scenario to evaluate the efficiency of that treatment.

The calculations were done for 1 m², and for 1 m depth, resulting in a cube of 1 m³ soil. The only removal process considered is uptake of heavy metals by trees.

The underlying equations of the mass balance model are (Algreen et al. 2013):

$$BCF = C_{\text{plant}}/C_{\text{soil}}$$

where BCF is the bioconcentration factor in plants, C_{plant} is the measured concentration in the plant (mg/kg) and C_{soil} is the contaminant concentration in the soil (mg/kg).

The loss by phytoextraction (no degradation or wash out), i.e. the change of contaminant mass (mg) is

$$dm/dt = -C_{\text{plant}} * dM_{\text{plant}}/dt$$

where dM_{plant}/dt is the growth of plant mass with time (kg/year).

Concentration (mg/kg) is mass of pollutant per mass of medium (kg), here soil:

$$dC_{\text{soil}}/dt = dm/(dt M_{\text{soil}})$$

The first into the second into the third equation gives (Trapp 2004):

$$dC_{\text{soil}}/dt = -BCF * C_{\text{soil}} * (dM_{\text{plant}}/dt)/M_{\text{soil}} = -k * C_{\text{soil}}$$

where k is the overall removal rate (year⁻¹). The analytical solution of this differential equation is (Trapp and Matthies 1998)

$$C_{\text{soil}}(t) = C_{\text{soil}}(0) * e^{-kt}$$

where $C_{\text{soil}}(0)$ is the initial soil concentration (mg/kg) and $C_{\text{soil}}(t)$ is the final target concentration (mg/kg). The time to reach this target concentration is then

$$t = \ln(C_{\text{soil}}(0)/C_{\text{soil}}(t))/k$$

Typical values for soil were used to parameterize the model, see Table 8.5. The value for soil and plant is a pair of concentrations measured nearby. The concentration of Cd (90 mg/kg) is the highest of all measured values. It can be seen that the calculation leads to absurd long removal times (12 million years). The Cd concentration measured in this campaign is with 90 mg/kg higher than the earlier value shown in Tables 8.1 and 8.4. But also with the median concentration for Cd, 2.8 mg/kg, and unchanged concentration in wood, the removal time would take >80 000 years. It can be concluded that a removal of heavy metals in Hunedoara by uptake into tree wood is an ineffective process, due to the low BCF-values.

Table 8.5 Parameterization of mass balance model

C_{soil} mg/kg	C_{tree} mg/kg	BCF kg/kg	Density ρ_{soil} kg/m ³	Plant growth kg/year/m ²	Loss rate k year ⁻¹	Target C_{soil} mg/kg	Time t year
90	0.0426	0.00047	1600	1	3.6×10^{-7}	1	12358632

8.2 Conclusions Hunedoara

We can then move on to flow chart 2 - and find no applicable phytoremediation method: there is no root zone degradation of heavy metals, the plants do not take up relevant amounts of heavy metals, and neither rhizo/phytodegradation, nor phytovolatilization, nor phytoextraction are an option.

Phytoremediation is not a feasible method for cleaning of the site at Hunedoara.

9. Evaluation of phytoremediation at the Szprotawa site

The same principles, methods, data and models can be used to judge the feasibility of phytoremediation at the Timbre site Szprotawa, Poland.

9.1 Flow chart Szprotawa

The flow chart shown in section 4.1 is applied for the situation in Szprotawa

Step 1: Do usage of the area prohibit plant growth?

The future land use of the area does not prohibit usage as forest or park area, thus, the answer is no.

Step 2: Is time of essence?

No.

Step 3: Are the conditions at site toxic to plants?

a) Inspection of plant growth at site.

At Szprotawa, a spontaneous pioneer vegetation developed that is composed of grasslands and trees such as poplars, willows, pine and birch. No signs of toxicity to plants were noticed.

b) Measured concentrations compared to toxicity tests

The concentrations measured at Szprotawa can be compared to toxicity data from controlled studies. It has to be considered that most of the contamination at Szprotawa concerns mainly the depth near or in the groundwater, in a depth of 1.5 to 1.9 m, and that this leads to a reduction of toxicity.

Table 9.1 shows the measured concentrations in soil at Szprotawa in 1 m and in 2 m depth. The difference is quite significant: sum BTEX is factor 55 (maximum values) to 18 (median) higher in 2 m depth than in 1 m depth, sum light alkanes factor 97 (maximum) and 187 (median).

The measured concentrations can be compared to the measured toxicity thresholds obtained by the tree toxicity testing (section 6.1). Toxic levels are seen in 2 m depth only, for the sum of light alkanes. Both in tests with spiked gasoline (super, ROZ 95) and with weathered contaminated soil, laboratory and in field studies, toxic effects of light gasoline ("Superbenzin", ROZ 95) of willow trees has been observed. Trees react to toxicity in deep soil by developing less (if any) roots in that layer. If a sufficient layer of clean top soil is available, trees can nonetheless grow, unless drought appears. It can thus be concluded that levels of hydrocarbons (BTEX, fuel) in Szprotawa are not lethal to trees but may inhibit growth in dry years.

Table 9.1 Measured concentrations (mg/kg dw) in soil Szprotawa, Poland, contrasted to EC50 data from the tree toxicity test (Table 5.1a-e).

1 m depth (mg/kg)	Max	Mean	Median	EC 50
Sum BTEX	4.3	2.3	2.3	292 (benzene)
Sum light gasoline	115.0	11.9	6.2	< 1000
2 m depth (GW level) (mg/kg)	Max	Mean	Median	
Benzene	1.7	0.3	0.06	292
Toluene	10.5	1.9	0.8	< 500
Ethylbenzene	79.0	14.2	2.9	nd
Xylenes	160	39	34	nd
Sum BTEX	240	55	41	benzene: 292
Sum light gasoline	11145	2310	1160	< 1000

Step 4. Degradation rates

Under aerobic or at least oxic conditions, short- and long-chain hydrocarbons (alkanes) and BTEX are quickly degraded (see section 8 for a description). The spill in Szprotawa is more than ten years old. Nitrates, reducible irons and sulphate have therefore very likely already been used by bacteria and are no more available as electron acceptors. Degradation is then limited by the downwards transport and availability of new electron acceptors - oxygen from air, and the "immediate reaction model" is used to quantify the kinetics of biodegradation (Wiedemeier et al. 1999). According to Wiedemeier, the loss rates of BTEX and light alkanes are similar (while those of long-chain alkanes would be slower). On the other hand, concentrations of (sum of) light gasoline (primarily light alkanes and derivatives) are about 100 times higher than sum of BTEX. Thus, calculations were made for light alkanes. About 3.5 g O₂ are needed to oxidise 1 g octane, which is used here as model compound for light alkanes.

The groundwater level in Szprotawa is 2 m depth, i.e. twice as deep as in the example calculation made in section 7. On the other hand, Neale et al. (2000) showed that the oxygen flux downwards in sandy soil does not much decrease with depth from 1 m to 2 m. We can thus use the result obtained in section 7, which is 11 g O₂/m²/d. A second loss of alkanes and BTEX is the uptake by trees, and this can be calculated from the transpiration of water (Trapp 2007).

Step 5. Volatilization

Doesn't apply, the pollution is in GW

Step 6. Mass balance model Szprotawa

The two dominant contaminant mass loss processes are loss by plant uptake and the loss by biological oxidation.

$$dm/dt = \text{deg} + \text{tree}$$

Where dm/dt is the change of mass with time (mg/a), deg indicates the degradation of the contaminants (mg/a) and tree is the mass loss by plant uptake (mg/a).

$$\text{deg} = J_O \cdot A/n$$

Where J_O is the diffusive flux of oxygen to the subsoil (mg/m²/a), A is the area (m²) and n is the number of mole oxygen required to oxidize 1 mole of contaminant (3.5 mol oxygen per mol alkanes).

$$\text{tree} = C_w \cdot Q/A$$

Where C_w is the concentration of contaminant (jet fuel) in groundwater (mg/l) and Q is the water flux taken up by plants (L/m²/a).

Trees have a second effect on the loss processes: Additional oxygen transport downwards will occur simply by the transpiration of water - water pores are replaced by gas pores, and a transpiration of 100 L water per year means an additional transport of 100 L air downwards, which contains then 30 g oxygen. Furthermore, the transpirations dries out the soil and is thus responsible for the presence of gas-filled pores, which enable 10 000 faster diffusion of gases than water-filled pores (see above, or Trapp and Matthies 1998).

An unknown value is the total concentration in groundwater: measured is the concentration dissolved in pore water, but there may also a fraction adsorbed, and there is at some places a non-aqueous phase liquid NAPL. Since jet fuel is less dense than water, this NAPL swims on top of the GW. The maximum concentration of light alkanes ("benzines") is 93 mg/L (Sep 2012, sample GWS52).

The K_d represents the ratio between (measured and available) concentration and thus mass in GW and (unknown) total concentration and mass (in dry soil or aquifer material) and is a key parameter for the mass balance. The maximum concentration in the deepest soil layer (2 m depth, sample S103) is 11145 mg/kg. A theoretical K_d is then 120 L/kg - which is very high for light alkanes in an aquifer and indicates the existence of a NAPL. The median values are GW 7.235 mg/L, soil 2 m depth 1160 mg/kg, giving a K_d of 160 L/kg. The respective values for benzene (the B in BTEX) are 0.073 (max) or 0.010 (median) mg/L in GW and 1.7 (0.06) mg/kg, giving a K_d of 23 L/kg. An estimated K_d of benzene would be at 0.45 L/kg (see Hunedoara). We can thus expect that the high K_d calculated from the measured dissolved concentration in GW and the measured total concentration in soil really are due to the existence of a NAPL or a fringe zone on top of the GW. Having clarified this, we set the K_d to a value of 1.0 L/kg. and treat the NAPL separately. The concentrations in soil were measured at 1 m and 2 m depth (see Table 9.1), thus, the high concentrations may appear maximally over a layer of 1 m thickness. The depth of the contaminated aquifer is set to 3 m (exact depth undetermined). A summary of input data is shown in Table 9.2

The mass balance calculations show that, as long as the soil is aerobic, and oxygen is transported down to NAPL and GW, the degradation of jet fuel proceeds rapidly. With 10 g O₂/d, about 3 g alkanes can be degraded, or, per year, 1043 g per m². This is sufficient to degrade the plume within a period of 11 years. Uptake by trees does only remove 9 g/year, which is less than 1% of the degradation. Interestingly, these results are in good agreement

with the prognosis made for the tank station in Axelved (Larsen et al. 2001). The experience from Axelved tells us that the model prognosis was too optimistic. This is because gasoline in GW was not degraded as fast - oxygen rarely and very slowly penetrates water-filled soil or aquifers (Braeckevelt et al. 2011), and gasoline is still present (14 years after the study was done) in the deeper layers at Axelved, which are water filled aquifers.

Transferring this knowledge to Szprotawa tells us that degradation is rapid as long as the soil is aerated. The plume will most likely not spread, and the NAPL will melt away. But deeper GW will not be clean as fast as predicted, because of transport limitations.

In conclusion, biodegradation is certainly a rapid and important loss process at Szprotawa, and trees support this loss process both by transpiring water from the soil, and by taking up some amount of pollutant.

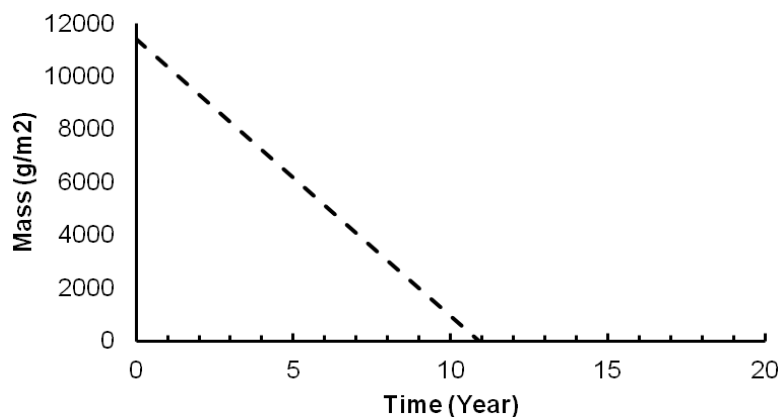


Figure 9.1 Simulated mass of jet fuel in Szprotawa (1-4 m depth) with maximum initial concentration, but optimistic (fully aerated) scenario.

Table 9.2 Input data for the mass balance model Szprotawa

Parameter	Value	Unit	Comment
Depth GW	3	m	unknown
Thickness NAPL	1	m	pessimistic assumption
Thickness NAPL	0.2	m	realistic
Initial C GW	93	mg/L	maximum
Initial C GW	7.235	mg/L	median
Initial C in NAPL layer	11145	mg/kg	soil 2 m depth maximum
Initial C in NAPL layer	1160	mg/kg	soil 2 m depth median
O ₂ -Flux	10	g/d	optimistic, aerated
O ₂ -flux	10×10^{-6}	g/d	pessimistic, stagnant water
Q water uptake trees	100	L/d	forest
Q water uptake trees	0	L/d	no vegetation

Having gone through flowchart 1 (Figure 4.1A) we can move on to flow chart Figure 4.1B for determination of possible (phyto)remediation processes and can identify here rhizodegradation, phytodegradation, hydraulic control and also phytovolatilization as potentially applicable processes at Szprotawa. From the mass balance calculations we know that rhizodegradation (degradation in or below the root zone) will be the most rapid and thus dominating process.

9.2 Conclusion for phytoremediation at Szprotawa

Phytoremediation is a feasible option at Szprotawa and will support the natural attenuation of the fuel products. The prognosis is that within one or few decades, jet fuel will vanish from top soil and deeper aerated soil (vadose zone), while rests and lenses of hydrocarbon will remain for much longer (several decades) in groundwater, but with shrinking extension.

9.3 Costs

The cost of phytoremediation greatly depends on monitoring activities that are required by authorities for any natural attenuation project. We will here only list the extra costs that appear due to the trees. In addition to the monitoring, additional expenses are expected due to planting of trees, for weeding, maintaining and (in case of drought or other calamities that lead to loss of many trees) replanting. Often willows and poplar are preferred to other tree species due to their high transpiration and fast growth (Timbre 2013b). Moreover, these species can be provided - and planted - as cuttings, which reduces the costs quite significantly. The costs per cutting can be down to $< 1/2$ € (Ny Vraa, 2013) and planting done by unlearned labour can be done for $< 10\,000$ €/ha (this depends, of course, on the minimum fee in the respective country. In Denmark, such low prizes can only be achieved exploiting children, and regular work of these is mostly illegal; but we once had the young pioneers engaged for such a project). In forestry it is common to plant initially denser, 4 to 8 trees per m^2 . If 1 cutting is needed per m^2 the price for 1 ha is app. 20 000 to 40 000 Euro costs for the trees. Alternatively, the usually upcoming natural pioneer vegetation can be supported, with much less costs, but slower appearance of a forest. Doing phytoextraction with trees allows harvesting of wood. Disposal is not necessary when treating degradable and volatile contamination; and the wood can perfectly be used and sold on the market. It is common to use the wood from such sites as biofuel (e.g., for pellets production, or for fire wood). Residuals of jet fuel would not damage such use but even lead to better material.

10. Discussion and recommendations for phytoremediation

When the phytoremediation technology arose the expectancies were high as research seemed promising. However, non-satisfactory field applications have often been reported (Gerhardt et al. 2009), leaving the phytoremediation technology with a less positive reputation. Despite of this, phytoremediation still has potential as a useful remediation method.

It is pivotal to emphasize the importance of site and contaminant characteristics. It is crucial to gather as much data concerning, physical chemical properties, total mass, toxicity and degradability of the pollution present, as possible in order to assess realistic remediation time. The predominant limitation of phytoremediation seems to be the long, sometimes extremely long, remediation time frame. Especially phytoextraction of heavy metals takes long time (thousands of years, predicted) and does not provide a final solution because the heavy metals reside in wood or even cycle between top soil and leaves or spread in the ecosystem. However, heavy metals cannot be destroyed, and there is no real treatment for them except making them non-available. This means dumping in deep deep holes, or fixation in glas or street sublayers. The volume of dump material can of course be reduced by methods such as soil washing or soil electrics, or burning of organic matter, but heavy metals as such will never be destroyed by these methods. On the other hand, there is no need for a site clean-up when the site is used for forestry purposes only - unless another good needs to be protected, such as groundwater. Allowing tree stands over centuries does not cost any maintenance and will, in most cases, be the cheapest alternative versus clean-up of any kind.

Contrary, in urban areas, where the site is to be used soon again, and where soil (or better: area) has a high value, a slow and long-lasting treatment method such as phytoremediation will not be a competitive treatment method. Alone the time frame will in many cases completely rule out phytoremediation. Unfortunately, most if not all (Western) cities that are older than 100 years have heavily polluted top soils. The reasons are heating with coal fires, traffic with leaded fuel and the non-existence of any rules for industrial production and waste before the late 1960s.

In Denmark, phytoremediation has been tested on several sites, and the results were considered as disappointment. From a scientific perspective, this is due to unrealistic expectations, or at least ill-designed projects. Some examples: We have investigated the Valby sludge basin (Algreen et al. 2013). There, a >5 m layer of heaviest polluted sludge (organic and inorganic) was planted with willows and trees. Due to methane production, rooting depth rarely exceeds 1 m, and plants suffered the first years, many died. After ten years, the content of some pollutants was measured again (Algreen et al. 2013), and no significant difference to the initial levels was found. We do not know what the engineers carrying out the project had expected. But it seems, from a scientific perspective, rather clear that a 5 m layer of anoxic toxic waste does not get valuable soil within a few years just by planting trees on top. Another well-investigated site is former gas works the Holte midpoint (Trapp and Christiansen 2003). There, cyanide is present as Prussian blue, which forms solid blue particles. In a first cleaning attempt, an engineering company tried to remove the cyanide substrate by excavation. Later investigations showed that at least ten times more than expected substrate remained. Poplar trees were planted in the year 1999/2000 and

grew very good at the site. The site owner (the local commune) was disappointed that cyanide (Prussian blue) was still present after ten years. Our scientific analysis showed that cyanide is very well degraded both as free cyanide (HCN and the anion) and as simple iron cyanide (ferro- and ferricyanide) (Trapp and Christiansen 2003, Larsen et al. 2005, Larsen and Trapp 2006), but the time-limiting factor for the cyanide removal is the dissolution kinetics of Prussian blue (ferriferrocyanide). Prussian blue is a solid that can of course neither be taken up by plants nor be attacked by bacteria. The dissolution depends mostly on pH and would have taken about 1000 years. This was known at the time of planting the trees. During dissolution, all free cyanide and all soluble iron cyanide in reach of the plants is rapidly degraded, making phytoremediation a protection measure against mobile cyanide. This effect is of course highly desired. But plants cannot be a treatment method for solid cyanide. After ten years, the treatment was considered a failure, as it seems due to ignorance of these well-known scientific facts. Disappointment about phytoremediation can thus be partly explained by expectations of miracles and by naive hopes (or promises) of those ignoring the biological, chemical and physical principles of our world.

Phytoremediation always falls short when dealing with pollution in deep aquifers. A treatment of such aquifers is necessary when they shall be used for drinking water purposes. This is the case in Denmark but not in all other countries. Often, clean water is available from cheaper sources (lakes, rivers), and often, the mineral content of deeper groundwater prohibits its usage for drinking water purposes, as it is the case in large parts of Asia with arsenic (Argos et al. 2010) and in volcanic areas with fluoride.

The slow uptake of heavy metals into plants bears also a chance: “phytoexclusion” means the use of plants, non-food crops or also strains of agricultural crops which do not take up (or very little) heavy metals (Dickinson et al. 2009). These plants can then be used even on soils with pollution above quality standards to produce biofuel or food. A popular and wide-spread example where this concept is helpful is “Urban Gardening” - all over the world, the population in the mega cities starts to grow their own food, not only in developing countries, but also in rich Western cities such as New York or Copenhagen.

A major strength of phytoremediation is the applicability at huge size areas and the self-sufficiency. The implementation costs are fairly inexpensive, and may be compensated by harvest products. After implementation the main expenses are connected to monitoring of the site, if this is required. The monitoring costs equal those of (monitored) natural attenuation MNA. From that perspective, phytoremediation combined with the production of biofuel or wood, is the treatment of choice wherever a fast removal of pollution is not feasible, or not necessary, where a reuse of the area is not expected within a lifetime, and where the sheer size of the area makes all other treatment efforts far too expensive. There are many of such sites, among them:

- the remote mega-sites stemming from former mining activities
- brown coal and black coal heaps
- old waste sites and larger sludge disposal sites.
- abandoned military areas (e.g., Szprotawa)
- diffusively contaminated abandoned brownfields in non-urban areas.

There are plenty of those sites in Europe. Also "wild" phytoremediation is helpful, which means the development of the climax vegetation on an undisturbed site.

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Annex: Contributors to the report and Disclaimer

Contributors

This report was written by Stefan Trapp, using input from Arno Rein, Mette Algreen, Lauge Peter Westergaard Clausen and partners of the TIMBRE project. Results from former projects have been used as cited. The report has been edited by Stephan Bartke (UFZ).

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